International Journal of Environmental Engineering and Management
Volume 1, Number 1 (2010), pp. 39-50

http://www.ripublication.com

Effect of Arsenic on Photosynthesis, Growth and its Accumulation in the Tissues of *Allium cepa* (Onion)

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Abstract

Arsenic is a well known carcinogenic element, that can harm not only human health but, plant and bacteria as well. Replicated experiments confirmed that, Arsenic accumulates in the different tissues in different parts of the plant and, adversely affects the growth and productivity of the plants. It is a threat for millions of population in terms of health and food security both. Therefore, a pot experiment was designed and conducted to investigate the effect of arsenic on photosynthetic pigments, Chlorophyll-a and –b, growth behavior, and its accumulation in the tissues of different parts of onion plants (*Allium cepa*). Test plants were subjected to pot experiment under natural conditions. Four pots were prepared to grow onion plants, irrigated with equal volume of different Arsenic solution ($NaAs_3$), 0.00 mg/l, 0.200mg/l, 0.600mg/l, and 0.800mg/l concentration with one pot for control respectively, throughout the experiments.

Both chlorophyll-a and -b contents in onion leaf increased significantly with the increase of water arsenic concentrations. The highest chlorophyll-a (0.004847/g) and chlorophyll-b (0.006528/g) contents were estimated in the onion leaf irrigated with 0.800mg/l of Arsenic whereas, in control plant it was lowest (chl-a 0.002363/ and chl-b 0.004092/g). A high positive correlation was observed between water arsenic (R^2= 0.897 and 0.963) & soil arsenic (R^2= 0.926 and 0.919) with chlorophyll-a and chlorophyll-b respectively. High positive correlation was also observed even for onion growth verses soil arsenic and water arsenic (R^2=0.994 and 0.968) and water Arsenic with leaf biomass (R^2=0.973) respectively.

However, no Arsenic accumulation was detected in the tissues of different parts of the onion plants suggesting that, arsenic ($NaAs_3$) influenced the biochemistry of photosynthesis which ultimately resulted in the increase of onion growth and yield. Onion plants can be cultivated in the area where Arsenic containing water is being utilized for irrigating crops but, a chain of in-vitro studies are required to understand the biochemistry and mechanism that influenced growth and productivity of the onion plants.

Keywords: Arsenic, Onion, Chlorophyll, Growth.

Introduction

A metalloid belongs to the group 15 of the periodic table now became a global threat, not only for the human health but plants and animals also. This 'Nector Poison', has been characterized as 'Hepato-toxic', 'Hemo-toxic', 'Geno-toxic' and now 'Phyto-toxic' also. Naturally occurring Arsenic, as a water quality issue in South Asia, began to attract international attention in the early decade of the nineties, when widespread chronic arsenic poisoning cases became apparent in Bangladesh and later in West Bengal India[1-2]. Natural arsenic contamination of drinking water has been reported from over 70 countries world-wide, affecting an estimated 150 million people[3]. About 50 million of these people live in Bangladesh, 30 million in India and 33 million in six other countries of south and south-east Asia.

Bihar Gangetic Plain has an Agricultural based economy where, Arsenic was already been detected in hand tube wells used for irrigation[3]. Due to high cost involved in supplying Arsenic free water for irrigation purposes, communities were forced to use Arsenic contaminated hand tube wells or borings for irrigating crops and vegetables. It was recently been recognized that, arsenic-contaminated groundwater used for irrigation poses a serious health hazard to people feeded from the crops irrigated, and that arsenic also accumulated in irrigated soils poses a serious threat to sustainable agriculture in affected areas[4]. Little is yet known about the extent and severity of this threat to food production, human health and livelihoods and rice, the staple food of the country, the principal crop affected. Therefore, a rigorous field and lab studies are required[5].

Arsenic content was found in variety of foods. The highest levels were detected in seafood, meats, and grains. High concentrations of as also occur in rice, mushrooms, poultry; root crops such as carrots, onions and potatoes are the most vulnerable ones. Considering plants, Marin et al. (1993) stated that, at higher concentration, arsenic was toxic to most plants. It interfered with metabolic processes and inhibited plant growth and development through arsenic induced phytotoxicity[6]. Later, Marin et al.(1992), Carbonell et al., (1995), Abedin et al. (2002a,2002b), Johan et al. (2003) has confirmed that, when plants were exposed to excess arsenic either in soil or in solution culture, they exhibited toxicity symptoms such as: inhibition of seed germination[7] , decrease in plant height[8-11], depress in tillering[12], reduction in root growth[7], decrease in shoot growth[13], lower fruit and grain yield[9,11,14] , and sometimes, leads to death[7,14]. However, little is known about the effect of arsenic on photosynthesis; the basis of plant bio-chemical system[15].

Majority of study covered and analyzed the adverse physiological and agronomical effects of arsenic related to the basic photochemical reaction so, the present research emphasized and analyzed chlorophyll pigments (a & b); the major photosynthetic pigments, in onion leaf to know the effect of Arsenic on the growth, yield and its accumulation[16].

Onions is a common vegetable for local communities as they used it as their staple food. They use raw onion as salad etc. Apart from onion bulb, onion leaves are also used in different ways as food sources. Thus, onion plant was selected for this study. In addition to that it was also easy to cultivate, monitor and harvest.

Materials and Methods

Pot experiment was conducted in natural conditions and the duration of the experiment was about 45 days from transplanting to harvest. All the pots were filled with similar soil. Healthy onion bulbs of similar size were grown in the different pots, i.e., control and other pots irrigated with equal volume of Arsenic solution ($NaAs_3$) of 0.200mg/l, 0.600mg/l, and 0.800mg/l respectively throughout the experiments.

Soil and Pot preparation

Soil was collected from a local nursey 'Pankaj Park, managed by Tarumitra(NGO),Patna and three kg of soil was taken in each of a series of 4 liter of volumetric pots. The unwanted materials such as dry roots, grasses, stones were removed and the soil was mixed thoroughly. Soil samples were tested before implanting the onion bulbs for moisture content, pH, Organic matter, & Arsenic etc. and then healthy onion bulbs of similar sized was grown in the different pots (Fig-1).

Soil Arsenic treatment

For the Arsenic treatment of soil, Sodium Meta Arsenate ($NaAs_2$) was used as the source of Arsenic. Each pot was irrigated by 200 ml each of different concentration of Arsenic solution 0.00 mg/l, 0.200 mg/l, 0.600 mg/l and 0.800 mg/l respectively for 7 days. One pot was treated with only distilled water (0.00mg/l of As) as control. This was followed by subsequent treatment of 150 ml of Arsenic solution for each of the pots throughout the experiments. The background soil Arsenic level was 0.013 mg/kg and at the end of the experiment soil was analyzed to know the soil arsenic level.

Determination of photosynthetic pigments

Chlorophyll-a and -b contents in onion leaf were determined at the early matured stage spectrophotometrically. Leaves were cut into small pieces and 100 mg of the leaf was taken into a mortar to grind them finely by pestle with 5 ml of 80% acetone. The extract was centrifuged for about 20 minutes. The supernatant was transferred to a measuring cylinder and the process was repeated thrice to ensure that the supernatant was fully extracted and then the final volume was made up to 15 ml. by adding required volume of 80% Acetone. The optical density of each solution was measured at 663 and 645 nm against 80% acetone blank in 1.5 cm cell following the method of Arnon (1949).

Determination of plant growth and yield

Plant height and leaf biomass were estimated for the measurement of plant growth. Plant height was recorded at the day of harvesting using meter scale (Fig-2).

Determination of Arsenic in Soil

Five grams of oven dried soil sample was taken in a 100-ml conical flask and 50 ml of 0.5M $NaHCO_3$ solution was added. Then, content was shaken for 1 h in a “to and fro” horizontal shaker and after completion of shaking; the suspension was filtered through Whatman filter paper No. 42. The filtrate was collected for arsenic analysis with Spectrophotometer by standard SDDTC method.

Results and Discussions

Effects of arsenic on chlorophyll content in onion leaf -

Photosynthesis is the most important biochemical process of this living world. It serves as the world’s largest solar battery and converts massive amount of sunlight into electrical and then chemical energy[18]. The most important photosynthetic pigment is chloroplast, consists of two types of chlorophylls, chlorophyll-a and chlorophyll-b. Chemically they differ from each other and absorb light of different wavelength to perform photosynthesis.

Although there was no report on the effect of arsenic on chlorophyll content in onion plant, Miteva and Merakchiyska (2002) reported that, arsenic concentrations of 25 mg kg-1 soil did not have negative effect on the photosynthetic process in bean plants (Phaseolus vulgaris L.), while the higher doses (50 and 100 mg of As kg-1 soil) inhibited the photosynthesis by 42 and 32%, respectively[19].

The current experiment was conducted with onion plant (*Allium cepa*) and both chlorophyll-a and -b contents in onion leaf increased significantly with the increase of water Arsenic & soil arsenic concentrations (Table-1). A very high positive correlation of water & soil arsenic concentrations was found with chlrophyll-a (R^2 = 0.897 & 0.963) [Graph-1-(A), (B)] and chlorophyll-b (R^2 = 0.926 & 0.919) respectively [Graph-1 (C),(D)].

Growth of the onion plants were significantly increased with the increasing concentration of Arsenic in irrigated water and soil (Graph-2 and Graph-3). The mean value of height of onion plant was 32cm (n=4) with minimum (18 cm.) in control plant and maximum (45cm.) in plant treated with Arsenic laced water (0.080mg/l) (Table: 2). A very high positive correlation of water and soil arsenic with height of the onion plants (R^2= 0.968 & 0.994) were found (Graph-4 and Graph-5).

The leaf biomass was also increased with the increasing Arsenic concentration in water and soil (Graph-6 and Graph-7). The mean value of leaf biomass was 0.825g/dry wt. (n=4) (Table: 3). Here also a very high positive correlation between Arsenic content in water and soil with leaf biomass was observed (R^2 = 0.973) (Graph-8 and Graph-9).

Table 1: Chlorophyll content in different onion leaves treated with different As solution.

Pot No.	Arsenic in Irrigated Water (mg/l)	Arsenic in Soil (mg/kg)	Chl a/gm	Chl b/gm	Total Chlorophyll
1	0.00	0.013	0.002363	0.004092	0.00690498
2	0.200	0.0287	0.003707	0.004746	0.00900882
3	0.600	0.0445	0.004767	0.006426	0.01193679
4	0.800	0.0574	0.004847	0.006528	0.01209684

Table 2: Height of the mature onion plants treated with different concentration of Arsenic.

Pot No.	Arsenic in Irrigation Water (mg/l)	Arsenic in Soil (mg/kg)	Height (cm)
1	0.000	0.013	18
2	0.200	0.0287	28
3	0.600	0.0445	36
4	0.800	0.0574	45

Table 3: Leaf Biomass of mature onion plants treated with different As solution.

Pot No.	Arsenic in Irrigation Water (mg/l)	Arsenic in Soil (mg/kg)	Leaf Biomass (g/pot dry wt.)
1	0.000	0.013	0.600
2	0.200	0.0287	0.800
3	0.600	0.0445	0.900
4	0.800	0.0574	1.000

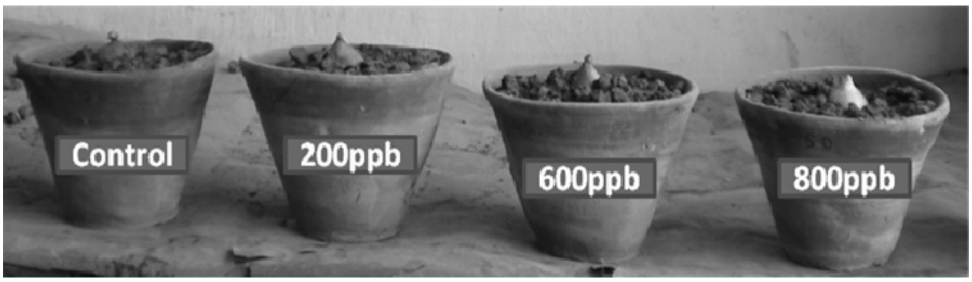


Figure 1: Cultivation of Onion bulb in different pot irrigated with different concentration of Arsenic.

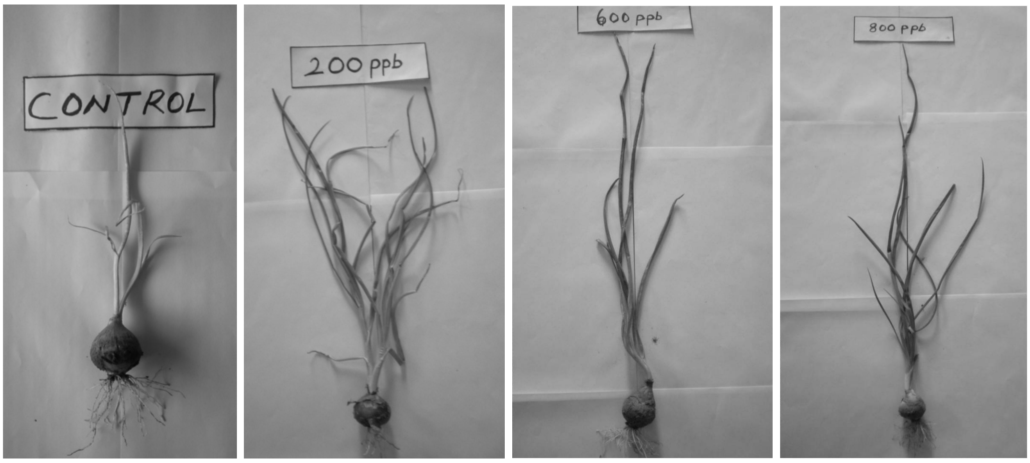
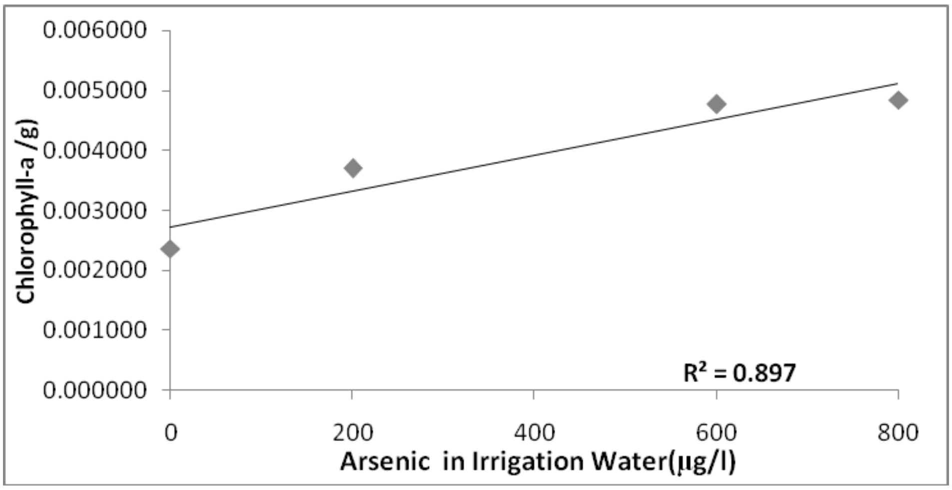
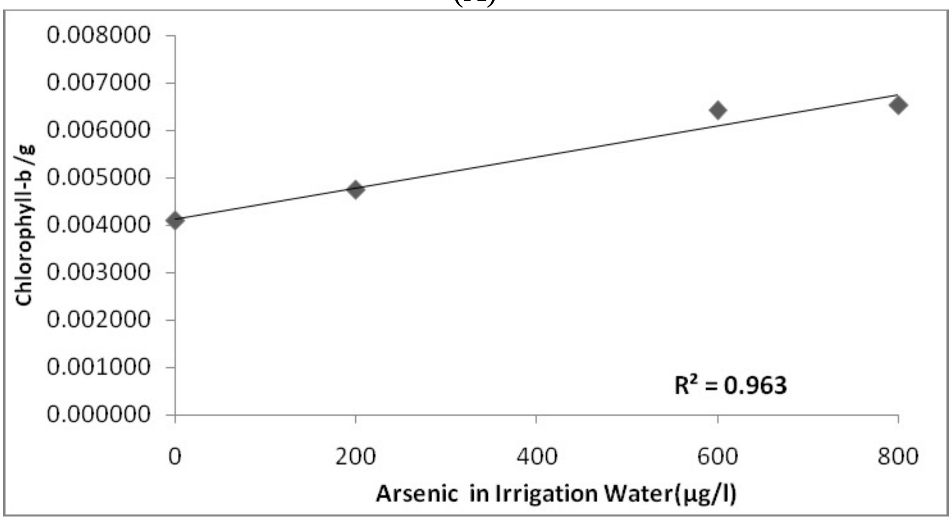


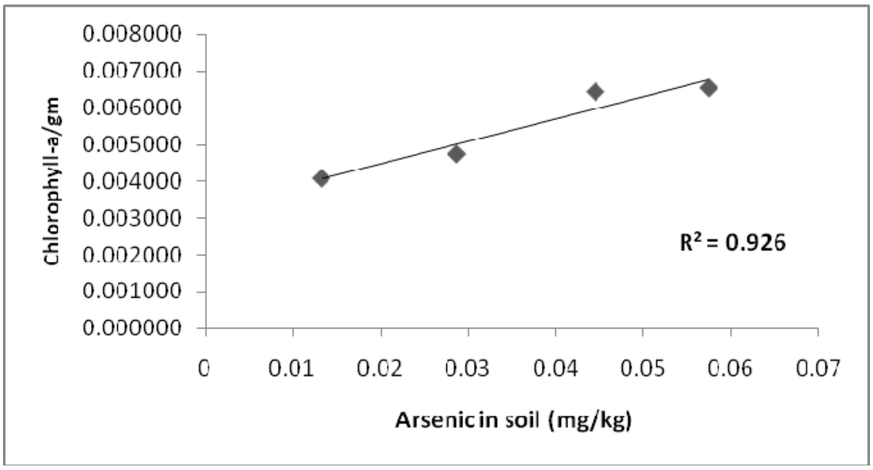
Figure 2: Matured harvested onion plants.



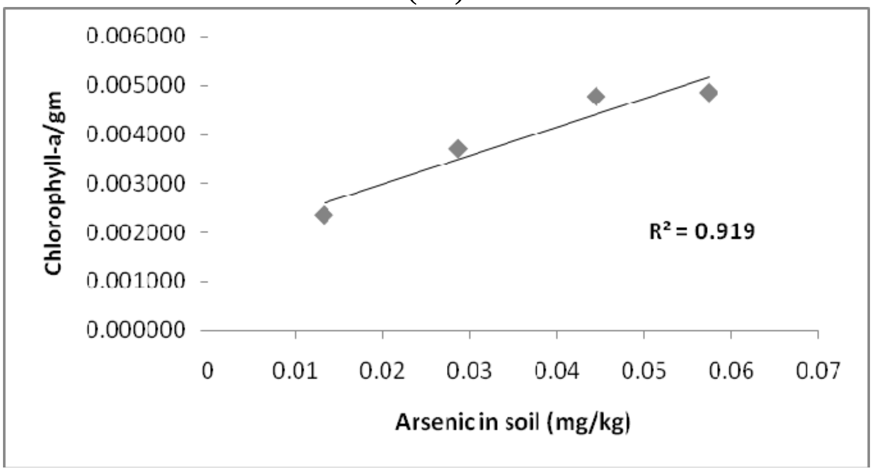
(A)



(B)

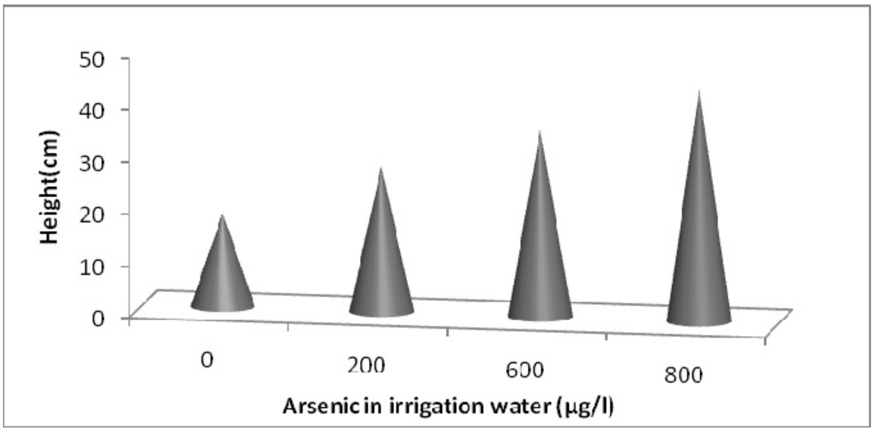


(C)

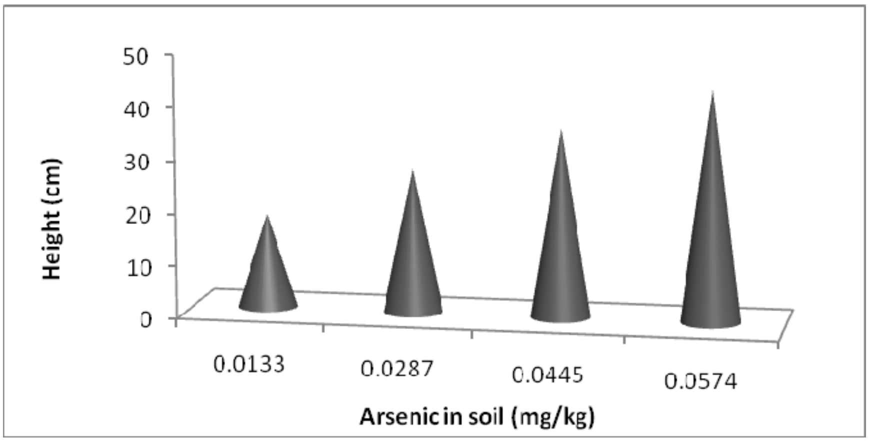


(D)

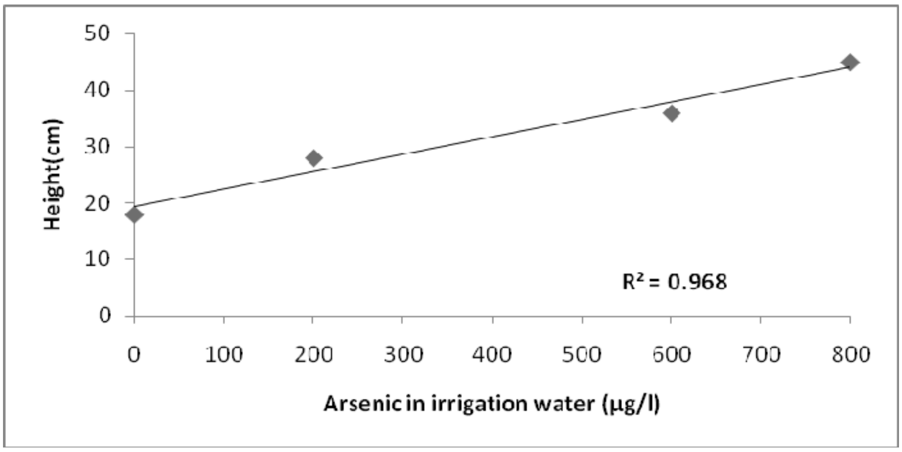
Graph 1: Correlation between Arsenic concentrations and photosynthetic pigments in onion leaves, chlorophyll-a (A, C) and chlorophyll-b (B, D).



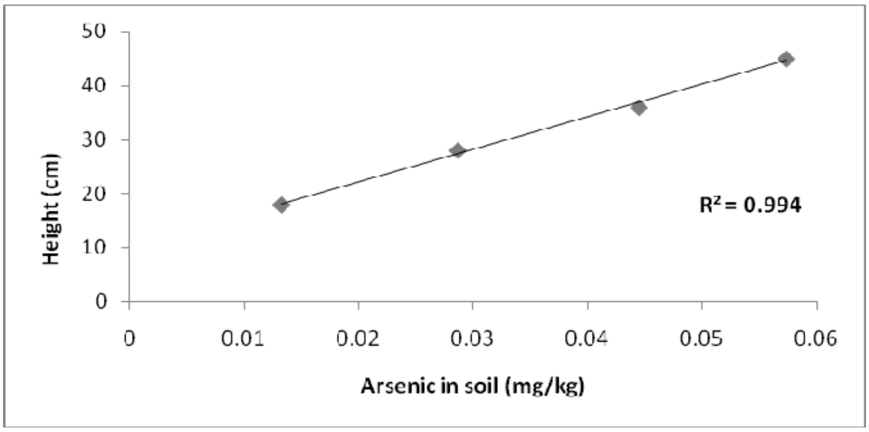
Graph 2: Height of the mature onion plants in different pots treated with different concentration of Arsenic.



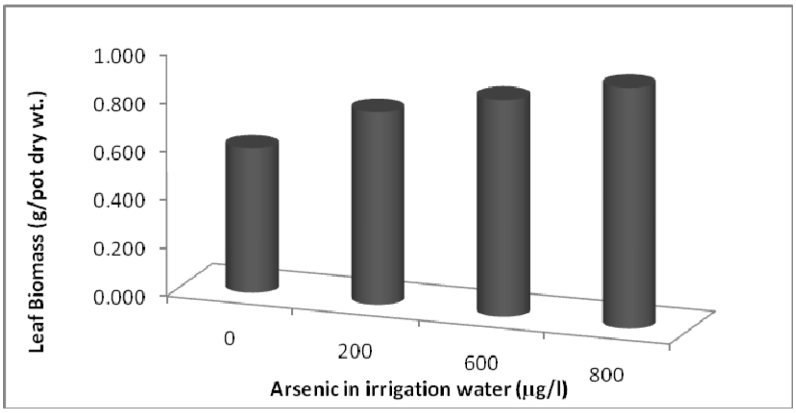
Graph 3: Height of the mature onion plants in different pots at different concentration of Arsenic in soil.



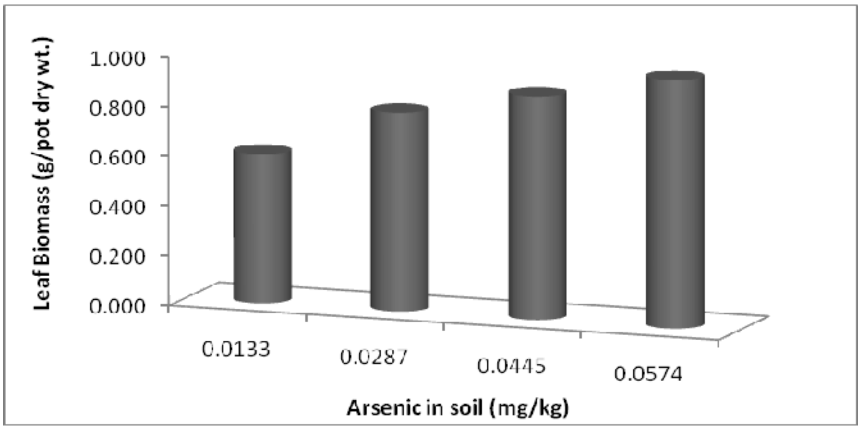
Graph 4: Correlation between Arsenic content in irrigation water and height of the plant.



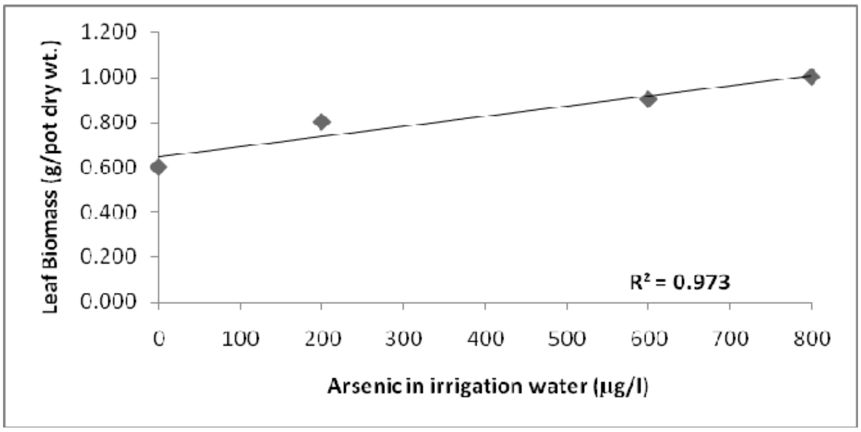
Graph 5: Correlation between Arsenic content in soil and height of the plant.



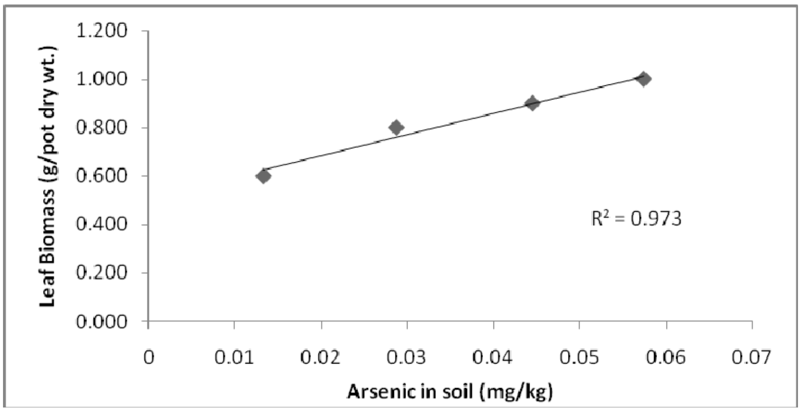
Graph 6: Leaf biomass of onion in different pots irrigated with different concentration of Arsenic.



Graph 7: Leaf biomass of onion in different pots at different concentration of Arsenic in soil.



Graph 8: Correlation between Arsenic content in water and leaf biomass of onion plants.



Graph 9: Correlation between Arsenic content in soil and leaf biomass of onion plants.

Conclusion

Arsenic was known for its toxic effects on plants, animals, chromosomes, genes etc. In plants it adversely affects the growth and yield of the plant. However, on the basis of observation and well correlations between chlorophyll content, growth and yield of onion, it may conclude that, Arsenic influenced the entire physiology positively and increased chlorophyll content significantly and geometrically in the onion leaves and thus, enhanced the growth and yield of onion. In this study Arsenic played a positive role as a growth promoter rather than growth inhibitor. Therefore, cultivation of onion (*Allium cepa*) plant on the area where Arsenic contaminated water is being utilized for irrigation can be done.

However, a chain of in-vitro study is required to know the biochemistry of the entire phenomena and to reveal the mechanism behind the positive and non toxic effect of Arsenic on the onion plant. This could be the future research.

Acknowledgment

The current study was part of my doctoral studies therefore, my special thanks to 'JAWAHARLAL NEHRU MEMORIAL FUND', New Delhi for funding my Ph.D. research work. I am also grateful to Mr. Anand Kumar, student of the Department of Environment and Water Management, A. N. College, Patna & Mrs. Rashmi Singh (sister) for their full cooperation and support during pot experiments. I am very much thankful to Ms. Aparupa Sengupta, Ph.D. student, MTU,USA for her intellectual inputs and valuable suggestions during editing the paper.

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